IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of	)	
	)	
ATSUMI et al	)	
	)	Art Unit: 122
Serial No.: 769,746	)	
	)	Examiner: Donald G. Daus
Filed: August 27, 1985	)	
	)	
For: NEW CEPHALOSPORIN COMPOUNDS AND THE PRODUCTION THEREOF	)	

DECLARATION UNDER 37 CFR 1.132

Honorable Commissioner of Patents and Trademarks

Washington, D.C.

Sir:

KUNIO ATSUMI, being duly warned, deposes and says that:

He is a citizen of Japan, residing at No. 3-16-11, Hiyoshi, Kohoku-ku, Yokohama-shi, Kanagawa-ken, Japan.

He is by profession a chemist, graduated Graduate School of Science and Technology, University of Tokyo Institute of Technology in March, 1979 and having received the degree of Ph. D. from University of Tokyo Institute of Technology.

Since April, 1979, he has worked for Meiji Seika Kaisha, Ltd. as an organic chemist working principally in the field of cephalosporins and related antibiotics.

He is the author or co-author of some 15 publications and an inventor or co-inventor of 8 pending

Japanese patent application and several foreign patent applications.

He is a member of the following technical and scientific societies:

Chemical Society of Japan.

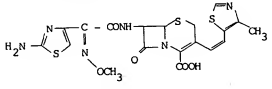
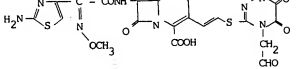
Pharmaceutical Society of Japan.

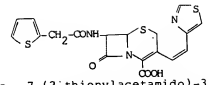
He has read the Office Action dated February 26, 1986 in the subject U.S. patent application, of which he is a co-inventor, as well as the references cited therein.

In order to distinguish the antibacterial activity possessed by the compounds of the present invention over the known similar cephalosporins of U.S. patent No. 4,307,116 of Farge et al (the cited reference B) which are similar to the compounds of the present invention in respect of the nature of the 7-position substituent but are different from the compounds of the present invention in respect of the nature of the substituent on the 3-position thiovinyl group, as well as over the known cephalosporins of U.S. patent No. 4,255,423 of Beattie et al (the cited reference A) which are similar to the compounds of the present invention in respect of the nature of the substituted vinyl group at the 3-position but are different from the compounds of the present invention in respect of the nature of the 7-position substituent, the following comparisons were conducted between them by determining

the values of the minimum inhibitory concentrations (MIC) against gram-positive bacteria and gram-negative bacteria of the compounds listed in the following Table 1.

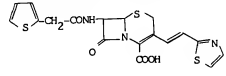
Table 1

Test Compounds	Nomination and Structure
Compound A according to the present invention	7-[2-methoxyimino-2-(2-amino-thiazol-4-yl) acetamido]-3-[2-(4-methylthiazol-5-yl) vinyl]-3-cephem-4-carboxylic acid (syn-isomer, (Z)-isomer, i.e. cis-isomer) of the formula 
Compound B (Comparative, according to U.S. patent No. 4,307,116 of Farge et al)	7-[2-methoxyimino-2-(2-amino thiazol-4-yl)acetamido]-3-[2-[5,6-dioxo-4-(2-formylmethyl)-1,4,5,6-tetrahydro-1,2,4-triazin-3-yl]thiovinyl]-3-cephem-4-carboxylic acid (syn-isomer, (E)-isomer, i.e. trans-isomer) of the formula 
Compound C (Comparative, according to a modification of U.S. patent No. 4,255,423 of Beattie et al)	7-(2-thienylacetamido)-3-[2-(thiazol-4-yl)vinyl]-3-cephem-4-carboxylic acid ((Z)-isomer, i.e. cis-isomer) of the formula



Compound D (Comparative, according to a modification of U.S. patent No. 4,255,423 of Beattie et al)

7-(2-thienylacetamido)-3-[2-(thiazol-2-yl)vinyl]-3-cephem-4-carboxylic acid ((E)-isomer, i.e. trans-isomer) of the formula



Compound A as above according to the present invention is representative of the compounds of the present invention and is the one as produced in Example 10 of the subject application and covered by the compound of the formula (If) according to the claim 7 and specifically set out in the claim 12 of the subject application.

Compound B (comparative) as above is identical to 7-[2-(2-amino-thiazol-4-yl)-2-methoxyiminoacetamido]-2-carboxy-3-[2-(5,6-dioxo-4-formylmethyl-1,4,5,6-tetrahydro-1,2,4-triazin-3-yl)-thiovinyl]-8-oxo-5-thia-1-aza-bicyclo [4.2.0]-oct-2-ene (syn-isomer, E-form) set out in the claim 3 of the cited U.S. patent No. 4,307,116 of Farge et al and is being studied in the clinical tests by Rhone-Poulenc Industries under the name "Ceftiolene", so that Compound B may be deemed as a representative and best one amongst the compounds of the cited reference of Farge et al.

Compound C (comparative) as above is a cephalosporin compound which is similar to the compounds of the claim 1 of the cited U.S. patent No. 4,255,423 of Beattie et al in respect of the 2-thienylacetamido substituent at the 7-position but has been modified from the compounds according to the U.S. patent of Beattie et al so as to have the thiazol-4-yl group introduced at the terminal carbon atom of the vinyl substituent at the 3-position in place of the values of R_2 and R_3 as defined in the claim 1 of the U.S. patent of Beattie et al, and this modification has been made with an intention that the substituted vinyl group at the 3-position of said Compound C is rendered more analogous to that of the above Compound A of the present invention than such a compound which would have such values of R_2 and R_3 as defined in the claim 1 of the U.S. patent of Beattie et al.

Compound D (comparative) as above is a cephalosporin compound according to a similar, second modification of the invention of the U.S. patent No. 4,255,423 of Beattie et al and is similar to the compounds of the claim 1 of the U.S. patent of Beattie et al in respect of the 2-thienylacetamido substituent at the 7-position but has been modified from the compounds of Beattie et al so as to have the thiazol-2-yl group introduced at the terminal of the vinyl substituent at the 3-position in

place of such values of R_2 and R_3 as defined by Beattie et al.

The MIC. values of these Compounds A, B, C and D were determined according to a standard serial dilution method by incubating the test organisms in "Sensitivity disk of agar-N" (a product commercially available from Nissui Co., Ltd., Japan) as an incubation medium at 37°C for 20 hours (overnight). The antibacterial spectra (MIC.) of the tested compounds so determined are shown in Table 2 below

Table 2

	MIC. value (μ/ml)			
	Test compounds			
Test organisms	A	B	C	D
Gram-positive bacteria				
Staphylococcus aureus 606	0.78	3.13	0.78	3.13
Staphylococcus aureus 209P JC-1	0.20	3.13	0.10	0.20
Staphylococcus epidermidis ATCC 14990	0.39	3.13	0.20	0.78
Enterococcus faecalis W-75	0.20	0.39	3.13	0.78
Gram-negative bacteria				
Escherichia coli NIHJ JC-2	0.39	0.20	25	25
Klebsiella oxytoca F-0100	1.56	25	>100	>100
Salmonella typhimurium LT-2	0.39	3.13	12.5	12.5
Proteus vulgaris GN 76	0.10	0.10	100	100
Proteus vulgaris GN76/C-1	0.39	0.78	>100	>100
Providencia rettgeri GN624	3.13	1.56	>100	>100
Serratia marcescens No. 1	0.39	0.39	>100	>100
Pseudomonas aeruginosa M-0148	25	>100	>100	>100
Pseudomonas aeruginosa MB-3833	25	50	>100	>100

From the comparisons between the results of Table 2 above, it can be observed that Compound A of the present invention shows a remarkably improved antibacterial activity against the tested four species of gram-positive bacteria over Compound B (comparative), though Compound A of the present invention is comparable to Compound B in respect of their antibacterial activity against the tested nine species of the gram-negative bacteria, that Compound A of the present invention shows a remarkably higher activity against the tested nine species of the gram-negative bacteria, over Compounds C and D (comparative), though Compound A is comparable to Compounds C and D for the antibacterial activities against the tested four species of the gram-positive bacteria, and that Compound A of the present invention is able to exhibit remarkably high antibacterial activities against a wide range of bacterial species, including both the tested four species of the gram-positive bacteria and the tested nine species of the gram-negative bacteria, in contrast to the fact that all of Compounds B, C and D (comparative) are less active against either one of the gram-positive bacteria species and the gram-negative bacteria species. This reveals that Compound A of the present invention is an excellent antibacterial agent which is much more greatly valuable in therapeutic treatments of bacterial infections, either mixed or singly,

as compared to the comparative Compounds B, C and D tested.

The excellent characteristic that the representative Compound A tested as above according to the present invention is able to exhibit the remarkably high antibacterial activities against a wide range of the bacterial species, including both the gram-negative bacteria and the gram-positive bacteria is common to all the compounds of the present inventions, as has been demonstrated in Table 1 on page 28 of the specification of the subject application which shows the MIC values (μ/mℓ) of the particular compounds of Examples 10-16, 18, 21 and 30-34 of the subject application. That this excellent characteristic exhibited by the compounds of the present invention is remarkable and surprising in view of that the G.L. Dunn's article on pages 1-10 of the "Journal of Antimicrobial Chemotherapy" (1982) 10, Suppl. C. 1-10 (the cited reference R of Dunn) has stated on page 2 lines 15-17 to the effect that the third-generation cephalosporins, including Cefotaxime, Ceftizoxime, Cefmenoxime, Ceftriaxone, Ceftazidime, Cefoperazone and Moxalactam (all of these agents containing the aminothiazolyloximino substituent at the 7-position commonly to the compounds of the present invention) tend to be less active than cephalosporin compounds of the earlier, first and second generations against gram-positive bacteria, most notably Staphylococcus aureus, with reference to the comparative in-vitro activity data

(MIC) of Ceftizoxime (one of the third-generation), Cefamandole (one of the second-generation) and Cephalothin (one of the first-generation) as shown in Table II on page 2 of the cited reference of Dunn. It is therefore surprising that the representative Compound A as tested of the present invention and the other compounds of the present invention are able to exhibit not only the remarkably high antibacterial activity of the MIC value of 0.78 μ/mℓ or less especially against Staphylococcus aureus, one of the gram-positive bacteria, but also the remarkably high antibacterial activities of the MIC values of 0.39 μ/mℓ or less against many of the gram-negative bacteria species. These excellent antibacterial properties of the compounds of the present invention, especially represented by Compound A as tested above are not predictable from the teaching of the cited reference of Farge et al and the teaching of the cited reference of Beattie et al, either alone or even in combination, I consider.

In addition, the compounds of the present invention are more highly absorbable through the intestines of a living animal when orally administered in the form of their esters with an enzymatically cleavable alcohol, as compared to the compounds of the cited reference of Farge et al and the compounds of the cited reference of Beattie et al. In order to demonstrate this, I and associated biologists

repeated the experiments described on page 32 line 15 to page 34 line 15 of the specification of the subject application, with using the pivaloyloxymethyl esters of Compound A, C and D as prepared by us. In this connection, I may add that no pivaloyloxymethyl ester could be prepared from Compound B when using the same method of esterification as that employed for the preparation of the corresponding pivaloyloxymethyl esters of Compounds A, B and C, and that hence we could neither obtain nor test a pivaloyloxymethyl ester of Compound B.

We repeated the experiments by the test procedure and the test conditions exactly same as described in Test 1 of on pages 33-34 of the specification of the subject application. The test results obtained are shown in Table 3 below.

Table 3

Test Compound	Rate of recovery of the test compound (in the free carboxylic acid form) in urine (%)
Pivaloyloxymethyl ester of Compound A according to the present invention	20%
Pivaloyloxymethyl ester of Compound C (comparative)	5.1%
Pivaloyloxymethyl ester of Compound D (comparative)	2.3%

The higher the rate of recovery of the test compound in the urine, the more the test compound as orally given is

absorbable through the intestines of the animal treated and the more the antibacterial potency of the test compound is maintainable to a substantial extent in the body of the animal until it is excreted in the urine, without receiving a substantial degradation of the cephalosporin compound in vivo. From the results of Table 3 above, therefore, it is noticeable that Compound A of the present invention is remarkably superior to Compounds C and D as an orally administrable antibacterial agent in that Compound A is more absorbable into the body of the animal through the intestines and is able to give a higher potency of the test compound (Compound A) in the blood for a prolonged time and to achieve its antibacterial power, as compared to Compounds C and D (comparative), until it is excreted into the urine.

I consider that the higher absorbability through the intestines of the pivaloyloxymethyl ester of the tested Compound A and hence of the compounds of the present invention is not predictable from the teaching of the cited reference of Farge et al and the teaching of the cited references of Beattie et al, where no reference is made at all to an improvement in the absorbability of the orally administered cephalosporins through the intestines of the animal.

The undersigned declarant declares further that all statements made herein of his own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Done at Yokohama, Japan, this 5 day of Sept., 1986

KUNIO ATSUMI

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